

What Is Claimed Is:

1. A method of measuring a holoceruloplasmin concentration in a blood spot based on an absorbance standard curve obtained through an enzyme-linked immunosorbent assay using a holoceruloplasmin-specific polyclonal antibody and a
5 holoceruloplasmin-specific monoclonal antibody.
2. A method of measuring a holoceruloplasmin concentration in a blood spot based on an fluorescence standard curve obtained through a dissociation-enhanced time-resolved fluoroimmunoassay using a holoceruloplasmin-specific polyclonal antibody
10 and a holoceruloplasmin-specific monoclonal antibody.
3. The method of claim 1 or claim 2, wherein the blood spot is collected using a blood filter paper.
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4. The method of claim 1 or claim 2, wherein the ceruloplasmin-specific polyclonal antibody is manufactured from the serum that is obtained by a rabbit immunized by the purified human ceruloplasmin containing the holoceruloplasmin.
5. The method of claim 1 or claim 2, wherein the ceruloplasmin-specific monoclonal antibody is manufactured from a hybridoma cell line obtained through
20 fusion of mouse spleen cells with myeloma cells, selection and cultivation of the fused spleen cells to produce an monoclonal antibody, and the spleen cells were obtained from an immunized mouse with the purified ceruloplasmin containing the holoceruloplasmin.
- 25 6. The method of claim 1, wherein the absorbance standard curve according

to the enzyme-linked immunosorbent assay is drawn out by applying the ceruloplasmin-specific polyclonal antibody and the ceruloplasmin-specific monoclonal antibody conjugated with horseradish peroxidase, respectively to a standard blood spot and a control reference blood spot manufactured.

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7. The method of claim 6, wherein the enzyme-linked immnosorbent assay for drawing out the absorbance standard curve is based on a sandwich method using the ceruloplasmin-specific polyclonal antibody and the ceruloplasmin-specific monoclonal antibody conjugated with horseradish peroxidase, respectively.

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8. The method of claim 2, wherein the fluorescence standard curve according to the dissociation-enhanced time-resolved fluoroimmunoassay is drawn out by applying the ceruloplasmin-specific polyclonal antibody and the ceruloplasmin-specific monoclonal antibody conjugated with europium, respectively to a standard blood spot and a control reference blood spot manufactured.

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9. The method of claim 8, wherein the dissociation-enhanced time-resolved fluoroimmunoassay for drawing out the fluorescence standard curve is based on a sandwich ELISA method using the ceruloplasmin-specific polyclonal antibody and the ceruloplasmin-specific monoclonal antibody conjugated with europium, respectively.

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10. A method of measuring a holoceruloplasmin concentration in a blood spot according to an enzyme-linked immunosorbent assay, the method comprising the steps of:

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manufacturing a ceruloplasmin-specific polyclonal antibody;

manufacturing a ceruloplasmin-specific monoclonal antibody;
conjugating horseradish peroxidase on the ceruloplasmin-specific polyclonal antibody and the ceruloplasmin-specific monoclonal antibody;
manufacturing a standard blood spot and a control reference blood spot by
5 removing a ceruloplasmin from a blood and adding a purified ceruloplasmin solution containing a holoceruloplasmin at a constant concentration to the blood;
drawing out an absorbance standard curve based on the standard blood spot and the control reference blood spot using the ceruloplasmin-specific polyclonal antibody and the ceruloplasmin-specific monoclonal antibody; and
10 measuring a holoceruloplasmin concentration in a blood spot of a patient using the standard curve through an enzyme-linked immunosorbent assay.

11. A method of measuring a holoceruloplasmin concentration in a blood spot according to a dissociation-enhanced time-resolved fluoroimmunoassay, the method comprising the steps of:
manufacturing a ceruloplasmin-specific polyclonal antibody;
manufacturing a ceruloplasmin-specific monoclonal antibody;
conjugating with europium on the ceruloplasmin-specific polyclonal antibody and the ceruloplasmin-specific monoclonal antibody;
20 manufacturing a standard blood spot and a control reference blood spot by removing a ceruloplasmin from a blood and adding a purified ceruloplasmin solution containing a holoceruloplasmin at a constant concentration to the blood;
drawing out an fluorescence standard curve based on the standard blood spot and the control reference blood spot using the ceruloplasmin-specific polyclonal antibody and the ceruloplasmin-specific monoclonal antibody; and
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measuring a holoceruloplasmin concentration in a blood spot of a patient using the standard curve through a dissociation-enhanced time-resolved fluoroimmunoassay.

12. The method of claim 10 or 11, wherein the patient has Wilson's disease.

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13. The method of claim 10 or 11, wherein the step of removing the ceruloplasmin in the blood is made using a phosphate-buffered saline.

14. The method of claim 10 or 11, wherein the standard blood spot and the

10 control reference blood spot are manufactured by adding a ceruloplasmin with a known concentration to the blood without the ceruloplasmin.

15. The method of claim 10 or 11, wherein the known concentration of the ceruloplasmin has at least 3 different values.

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16. The method of claim 10 or 11, comprising a further step of screening an antibody for neutralizing an oxidase activity of the holoceruloplasmin at the step of manufacturing the ceruloplasmin-specific monoclonal antibody.

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17. The method of claim 10 or 11, comprising a further step of purifying the antibody after the step of manufacturing the ceruloplasmin-specific monoclonal antibody.

18. A Wilson's disease screening kit reagent, comprising a

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holoceruloplasmin-specific polyclonal antibody, a holoceruloplasmin-specific

monoclonal antibody, a standard blood spot, and a control reference blood spot.

19. A Wilson's disease screening kit, being characterized of measuring a holoceruloplasmin concentration in a blood spot based on an absorbance standard curve obtained through an enzyme-linked immunosorbent assay using a holoceruloplasmin-specific polyclonal antibody, a holoceruloplasmin-specific monoclonal antibody, a standard blood spot, and a control reference blood spot.

20. A Wilson's disease screening kit, being characterized of measuring a holoceruloplasmin concentration in a blood spot based on an fluorescence standard curve obtained through a dissociation-enhanced time-resolved fluoroimmunoassay using a holoceruloplasmin-specific polyclonal antibody, a holoceruloplasmin-specific monoclonal antibody, a standard blood spot, and a control reference blood spot.

15 21. A method of diagnosing Wilson's disease using the screening kit of claim
19 or 20.